# M-TriDAP

PGN-derived molecule; a NOD1/NOD2 ligand

Catalog # tlrl-mtd

For research use only

Version # 16F30-MM

## PRODUCT INFORMATION

**Content:**
- 1 mg M-TriDAP
- 1.5 ml endotoxin-free water

**Storage:**
- M-TriDAP is provided as a lyophilized powder and shipped at room temperature. Store at -20 °C.
- Upon resuspension, prepare aliquots of M-TriDAP and store at -20 °C.
- Product is stable for 1 year at -20 °C when properly stored. Avoid repeated freeze-thaw cycles.

**Quality control:**
- The biological activity has been validated using HEK-Blue™ NOD1 and HEK-Blue™ NOD2 cells.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

## DESCRIPTION

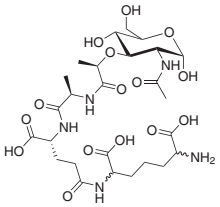
MurNAc-L-Ala-γ-D-Glu-mDAP (M-TriDAP), also called DAP-containing muramyl tripeptide is a peptidoglycan (PGN) degradation product found mostly in Gram-negative bacteria. M-TriDAP is recognized by the intracellular sensor NOD1 (CARD4) and to a lesser extent NOD2 (CARD15). Recognition of M-TriDAP by NOD1/NOD2 induces a signaling cascade involving the serine/threonine RIP2 (RICK, CARDIAK) kinase that interacts with IKK leading to the activation of NF-κB and the production of inflammatory cytokines such as TNF-α and IL-6[1]. M-TriDAP induces the activation of NF-κB at similar levels to Tri-DAP[2].

M-TriDAP provided by InvivoGen is chemically synthesized.
*Note: M-TriDAP is a mixture of MurNAc-L-Ala-γ-D-Glu-D-mDAP and MurNAc-L-Ala-γ-D-Glu-L-mDAP.*

**1. Park JH. et *al.*, 2007.** RICK/RIP2 mediates innate immune responses induced through Nod1 and Nod2 but not TLRs. J Immunol. 178(4):2380-6.
**2. Girardin SE. et al., 2003.** Peptidoglycan molecular requirements allowing detection by Nod1 and Nod2. J Biol Chem. 278(43):41702-8.

## CHEMICAL PROPERTIES

**Source:** Synthetic
**Synonym:** N-acetyl-muramyl-L-Ala-γ-D-Glu-meso-diaminopimelic acid
**Formula:** $C_{26}H_{43}N_5O_{15}$
**Molecular weight:** 665.64
**Structure:**



## METHODS

**Preparation of stock solution (1 mg/ml)**
- Add 1 ml endotoxin-free water (provided) and vortex until completely dissolved.

**Working concentrations**
- 1-10 µg/ml for the activation of NOD1
- 100 ng-10 µg/ml for the activation of NOD2

**NOD1/NOD2 activation using M-TriDAP**
M-TriDAP can be used to activate NOD1 or NOD2 in cells expressing these receptors, such as HEK-Blue™ NOD1 and HEK-Blue™ NOD2 cells. These cells express the human or mouse NOD1 or NOD2 genes and an NF-κB inducible SEAP reporter gene. Levels of SEAP can be easily determined using HEK−Blue™ Detection, a cell culture medium that allows the detection of SEAP as it is secreted by the cells.
For more information visit: www.invivogen.com

- Add 20 µl of M-TriDAP at various concentrations (100 ng-10 µg/ml) per well of a 96-well plate.
- Prepare a cell suspension (~280,000 cells per ml) in HEK-Blue™ Detection medium and immediately add 180 µl of the cell suspension (~50,000 cells) to each M-TriDAP-containing well.
- Incubate the plate for 6-24 h at 37 °C, 5% $CO_2$.
- Determine SEAP levels using a spectrophotometer at 620-655 nm.

## RELATED PRODUCTS

| Product | Cat. Code |
|---|---|
| HEK-Blue™ hNOD1 Cells (human NOD1 gene) | hkb-hnod1 |
| HEK-Blue™ hNOD2 Cells (human NOD2 gene) | hkb-hnod2 |
| HEK-Blue™ mNOD1 Cells (mouse NOD gene) | hkb-mnod1 |
| HEK-Blue™ mNOD2 Cells (mouse NOD2 gene) | hkb-mnod2 |

TECHNICAL SUPPORT
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